Bayer HealthCare

QC

CE 0088

HBc Total (HBcT)

Contents

REF	Contents
07569996	2 vials of Negative Control CONTROL
	2 vials of Positive Control CONTROL
	Expected Values Card and barcode labels

Preliminary 00367468 Rev. A, 2004-06

Intended Use

For *in vitro* diagnostic use in monitoring the performance of the HBc Total assay on the ADVIA Centaur® Systems. The performance of the HBc Total quality control material has not been established with any other anti-HBc Total assays.

Control Description

Volume	Ingredients	Storage	Stability
7.0 mL/vial	Processed human plasma negative and positive for anti-HBc with preservatives	2–8°C	Until the expiration date on the vial label or onboard–8 hours

R43 **Irritant!** May cause sensitization by skin contact. Avoid contact with skin.
S24, S37 Wear suitable gloves. **Contains:** 5-chloro-2-methyl-2H-isothiazol-3-one and 2-methyl-2H-isothiazol-3-one

CAUTION! POTENTIAL BIOHAZARD: The controls contain human source material. No known test method can offer complete assurance that products derived from human blood will not transmit infectious agents. All products manufactured using human source material should be handled as potentially infectious. Handle this product according to established good laboratory practices and universal precautions.[1-3] Use eye protection and gloves when handling this product; wash hands after handling.

The negative control has been assayed by FDA-approved methods and found nonreactive for hepatitis B surface antigen (HBsAg), antibody to hepatitis C (HCV), and antibody to HIV-1/2. The positive control contains human plasma that may be reactive for HBsAg. The units were treated with a BPL-UV inactivation procedure, however, all products manufactured using human source material should be handled as potentially infectious.

For *In Vitro* Diagnostic Use.

Preparing the Quality Control Material

Gently swirl and invert the vials to ensure homogeneity.

Using the Barcode Labels

NOTE: Control barcode labels are lot number specific. Do not use barcode labels from one lot of controls with any other lot of controls.

Use the HBc Total quality control barcode labels to identify the positive and negative sample cups when performing the ADVIA Centaur HBc Total assay. Place the barcode label on the sample cup so that the readable characters on the side of the label are vertical on the sample cup.

Performing Quality Control

For detailed information about entering quality control values, refer to the system operating instructions or to the online help system.

To monitor system performance and chart trends, as a minimum requirement, quality control samples should be assayed on each workshift that samples are analyzed. Quality control samples should also be assayed when performing a two-point calibration. Treat all quality control samples the same as patient samples.

NOTE: This procedure uses control volumes sufficient to measure each control in duplicate.

1. Schedule the quality control samples to the worklist.
2. Label two sample cups with quality control barcode labels: one for the positive, and another for the negative.

NOTE: Each drop from the control vial is approximately 50 µL.

3. Gently mix the quality control materials and dispense at least 5 to 6 drops into the appropriate sample cups.
4. Load the sample cups in a rack.
5. Place the rack in the sample entry queue.
6. Ensure that the assay reagents are loaded.
7. Start the entry queue, if required.

NOTE: Dispose of any quality control materials remaining in the sample cups after 8 hours. Do not refill sample cups when the contents are depleted; if required, dispense fresh quality control materials.

Reviewing, Editing, and Printing Results

For detailed information about reviewing, editing, and printing quality control results, refer to the system operating instructions or to the online help system.

Expected Results

Refer to the *Expected Values* card for the assigned values specific for the lot number of the HBc Total quality control material. The expected values are traceable to the standardization of the HBc Total assay. For additional information, refer to the reagent instructions for use.

The expected values should be used only as a guide in evaluating performance. Since performance is subject to the design and condition of each instrument or reagent system, it is recommended that each laboratory establish its own expected values and acceptable limits. The mean values established should fall within the range specified in *Expected Values*. Individual results may fall outside the range.

Taking Corrective Action

If the quality control results do not fall within the suggested *Expected Values* or within the laboratory's established values, then do the following:

- consider the sample results invalid and repeat testing if controls are out of range
- review these instructions to ensure that the assay was performed according to the procedures recommended by Bayer HealthCare
- verify that the materials are not expired
- verify that required maintenance was performed
- if necessary contact Bayer HealthCare for more assistance

Limitations

The results obtained using the HBc Total quality control material depend on several factors. Erroneous results can occur from improper storage, inadequate mixing, or sample handling errors associated with system or assay procedures.

- Do not return any quality control materials back into the vials after testing because evaporation and contamination can occur, which may affect results.
- Dispose of any quality control material remaining in the sample cups after 8 hours.
- Do not refill sample cups when the contents are depleted. If required, dispense fresh quality control materials.

Disposal

Dispose of hazardous or biologically contaminated materials according to the practices of your institution. Discard all materials in a safe and acceptable manner, and in compliance with all federal, state, and local requirements.

Technical Assistance

For customer support, please contact your local technical support provider or distributor.

References

1. National Committee for Clinical Laboratory Standards. Procedures for the Handling and Processing of Blood Specimens; Approved guideline-2nd Edition. NCCLS document H18-A2. Wayne (PA):NCCLS;1999.
2. Centers for Disease Control. Update: Universal precautions for prevention of transmission of human immunodeficiency virus, hepatitis B virus and other bloodborne pathogens in healthcare settings. MMWR 1988;37:377-82, 387-8.
3. National Committee for Clinical Laboratory Standards. Protection of laboratory workers from instrument biohazards and infectious disease transmitted by blood, body fluids, and tissue; approved guideline. NCCLS Document M29-A2. Wayne (PA):NCCLS;2001.

HBc Total (HBcT)

Assay for the Detection of Total Antibodies to Hepatitis B Core Antigen

Assay Summary

Sample Type	*Serum, potassium EDTA plasma, lithium or sodium heparinized plasma*
Sample Volume	*50 µL*
Calibrator	*HBcT*

Contents

REF	Contents	Number of Tests
07566733	1 ReadyPack® primary reagent pack containing ADVIA Centaur® HBcT Lite Reagent, Solid Phase, and Chaotrope Reagent	200
	1 Ancillary pack containing ADVIA Centaur HBcT Ancillary Reagent ANC	
	ADVIA Centaur HBcT Master Curve card	
	1 vial HBcT Low Calibrator CAL L	
	1 vial HBcT High Calibrator CAL N	
	ADVIA Centaur HBcT Calibrator Assigned Value card	

For a definition of symbols used in product labeling, please refer to Appendix D, *Understanding the Symbols*, in the *ADVIA Centaur® Assay Manual*.

Intended Use

The ADVIA Centaur HBc Total assay is an *in vitro* diagnostic test for the qualitative determination of total antibodies to the core antigen of the hepatitis B virus (HBc Total) in human serum or plasma (potassium EDTA, or lithium or sodium heparinized) using the ADVIA Centaur® System. This assay can be used as an aid in the diagnosis of individuals with acute or chronic hepatitis B virus (HBV) infection and in the determination of the clinical status of HBV infected individuals in conjunction with other HBV serological markers for the laboratory diagnosis of HBV disease associated with HBV infection. This assay can also be used as an aid in the differential diagnosis in individuals displaying signs and symptoms of hepatitis in whom etiology is unknown.

WARNING: This assay has not been FDA cleared or approved for the screening of blood or plasma donors.

United States federal law restricts this device to sale by or on the order of a physician.

Materials Required But Not Provided

REF	Description	Contents
	ADVIA Centaur System	
07569996	ADVIA Centaur HBcT quality control material	2 x 7.0 mL Negative Control CONTROL 2 x 7.0 mL Positive Control CONTROL Expected Value card
01137199 (112351)	ADVIA Centaur Wash 1 WASH	2 x 1500 mL/pack

Summary and Explanation of the Test

The ADVIA Centaur HBc Total assay is an antigen bridging microparticle chemiluminometric immunoassay used for the detection of antibodies to hepatitis B core antigen in human serum or plasma.

Hepatitis B virus (HBV) is endemic throughout the world and is the major cause of liver disease. HBV is transmitted through direct contact with blood and body fluids. Common modes of transmission include blood transfusion, needle puncture, direct contact with open wounds, sexual contact, and mother-neonate contact during birth.[1,2]The average incubation period for HBV infection is 6 to 8 weeks (range 1 to 6 months). Common clinical symptoms include malaise, fever, gastroenteritis, and icterus. HBV infection can result in typical icteric hepatitis, subclinical anicteric hepatitis, fulminant hepatitis, or chronic or persistent hepatitis. In adults, 90 to 95% of patients with HBV infection completely recover from acute illness and clear the virus. Approximately 5 to 10% of patients with HBV become chronic carriers. In HBV infected neonates, approximately 90% develop chronic hepatitis B infection. It is estimated that over 300 million people worldwide are chronic carriers of the virus. HBV infection, particularly in cases of chronic infection, is clearly associated with the development of hepatocellular carcinoma.[1,2,3]

Hepatitis B core antigen (HBcAg), found in liver cells, does not circulate in the bloodstream. However, IgM and IgG antibodies to HBcAg can be detected serologically in HBV infected individuals. Anti-HBc IgM is detectable first and remains detectable for approximately six months. Shortly after the IgM response, anti-HBc IgG appears and can remain detectable indefinitely. The presence of anti-HBc IgM and anti-HBc IgG is characteristic of acute infection, while the presence of anti-HBc IgG without anti-HBc IgM is characteristic of chronic or recovered stages of HBV infection. Anti-HBc Total assays detect both IgM and IgG anti-HBc responses. Most often levels of anti-HBc will coincide with detectable levels of other HBV markers. Rarely, anti-HBc may be the only detectable HBV marker. This may occur during the brief period when hepatitis B surface antigen (HBsAg) has been cleared from the bloodstream and before antibodies to hepatitis B surface antigen (anti-HBs) become detectable. For this reason, the use of anti-HBc Total assays to detect acute infection is not recommended. Anti-HBc Total assays should be used in conjunction with other marker assays to assess current or past exposure to HBV.[1,2,4,5]

Assay Principle

The ADVIA Centaur HBc Total assay is a two wash antigen sandwich immunoassay in which antigens are bridged by antibody present in the patient sample. The Solid Phase contains a preformed complex of streptavidin coated microparticles and biotinylated recombinant HBc antigen and is used to capture anti-HBc in the patient

sample. The Lite Reagent contains recombinant HBc antigen labeled with acridinium ester and is used to detect anti-HBc in the sample. Solid Phase and Chaotrope Reagent are added to the sample, followed by Ancillary Reagent and Lite Reagent. Antibody-antigen complexes will form if anti-HBc antibodies (IgM and IgG) are present in the sample.

The system automatically performs the following steps:

- dispenses 50 µL of sample into a cuvette and incubates for 6 minutes at 37°C
- dispenses 125 µL of Solid Phase and incubates for 18 minutes at 37°C
- dispenses 30 µL of Chaotrope Reagent (**NOTE:** The Chaotrope Reagent is colorless to light pink in color.)
- washes the cuvette with Wash 1
- dispenses 100 µL of Ancillary Reagent, incubates the mixture for 5.75 minutes at 37°C
- dispenses 50 µL of Lite Reagent, incubates the mixture for 18 minutes at 37°C
- separates the Solid Phase from the mixture and aspirates the unbound reagent
- washes the cuvette with Wash 1
- dispenses 300 µL each of Acid Reagent and Base Reagent to initiate the chemiluminescent reaction
- reports results according to the selected option, as described in the system operating instructions or in the online help system

The relative light units (RLUs) detected by the ADVIA Centaur System are used to calculate the Index Value from the Master Curve. Assay results above the cutoff of the assay are not indicative of antibody level. Refer to *Interpretation of Results* for a description of the Cutoff Value calculation.

Specimen Collection and Handling

Serum, potassium EDTA plasma, lithium or sodium heparinized plasma are the recommended sample types for this assay.

Heparin has been shown to decrease the Index values in some HBc Total reactive samples relative to serum (See under Alternative Sample Types). Results obtained from heparin specimens falling near the cutoff should be repeated with a serum specimen or interpreted with caution.

The Index values of anti-HBc reactive specimens that have been collected in Lithium Heparin and PST gel barrier tubes may be decreased or become non-reactive if these tubes are inverted prior to analysis. To avoid erroneous results samples must be re-centrifuged before analysis.

Do not use specimens with obvious microbial contamination. The performance of the ADVIA Centaur HBc Total assay has not been established with cord blood, neonatal specimens, cadaver specimens, heat-inactivated specimens, or body fluids other than serum or plasma such as saliva, urine, amniotic fluid, or pleural fluid.

The following general recommendations for handling and storing blood samples are furnished by the National Committee for Clinical Laboratory Standards[6], and augmented with additional sample handling studies using the ADVIA Centaur HBc Total assay:

- Handle all samples as if capable of transmitting disease.

- Samples are processed by centrifugation, typically followed by physical separation of the serum or plasma from the red cells. The centrifugation step may occur up to 24 hours post draw.
- Test samples as soon as possible after collecting. Store samples at 2 to 8°C if not tested within 12 hours of collection
- Store samples stoppered and upright at all times at 2 to 8°C up to 3 days.
- Freeze samples, devoid of red blood cells, at or below -20°C for longer storage. Do not store in a frost-free freezer. When 10 samples were subject to 2 freeze/thaw cycles, no clinically significant differences were observed. Thoroughly mix thawed samples and centrifuge at 10,000g for 2 min before using.
- Package and label samples for shipment in compliance with applicable federal and international regulations covering the transport of clinical samples and etiological agents. Samples maintained at room temperature up to 12 hours or refrigerated up to 3 days demonstrated no qualitative differences. Store samples stoppered and upright at 2 to 8°C upon arrival. If shipment is expected to exceed 3 days, ship specimens frozen.

Before placing samples on the system, ensure the following:

- Samples are free of fibrin or other particulate matter. Remove particulates by centrifugation (example: 1500xg for 10 minutes; follow tube manufacturer's recommendations[6])
- Samples are free of bubbles or foam.

Reagents

Store the reagents upright at 2–8°C.

Mix all primary reagent packs by hand before loading them onto the system. Visually inspect the bottom of the reagent pack to ensure that all particles are dispersed and resuspended. For detailed information about preparing the reagents for use, refer to Appendix C, *Handling Reagents*.

Protect from sunlight

Protect reagent packs from all light sources. Reagent packs loaded on the system are protected from light. Store unused reagent packs at 2–8°C away from light sources.

Reagent Pack	Reagent	Volume	Ingredients	Storage	Stability
ADVIA Centaur HBcT ReadyPack primary reagent pack	Lite Reagent	10.0 mL/ reagent pack	recombinant hepatitis B core antigen (~ 0.2 µg/mL) labeled with acridinium ester in buffer with surfactant and preservatives	2–8°C	until the expiration date on the pack label. For onboard stability, refer to *Onboard Stability and Calibration Interval*.
	Solid Phase	25.0 mL/ reagent pack	streptavidin coated paramagnetic microparticles preformed with biotinylated recombinant HBcAg (~ 0.2 µg/ mL) in buffer with potassium thiocyanate (5.0%), bovine serum albumin, surfactant, and sodium azide (< 0.1%)	2–8°C	until the expiration date on the pack label. For onboard stability, refer to *Onboard Stability and Calibration Interval*.

Reagent Pack	Reagent	Volume	Ingredients	Storage	Stability
	Chaotrope Reagent*	6.0 mL/ reagent pack	buffer with potassium thiocyanate (40%) and surfactant	2–8°C	until the expiration date on the pack label. For onboard stability, refer to *Onboard Stability and Calibration Interval.*
ADVIA Centaur HBcT ANC Ancillary Reagent Readypack	Ancillary Reagent	20.0 mL/ reagent pack	buffer with potassium thiocyanate (5.0%) and surfactant	2–8°C	until the expiration date on the pack label. For onboard stability, refer to *Onboard Stability and Calibration Interval.*
HBcT calibrator vials	Calibrators	2.0 mL/ vial	processed human plasma positive for HBc antibodies, bovine serum albumin and preservatives	2–8°C	until the expiration date on the vial or onboard 8 hours
HBcT quality control material vials**	Controls	7.0 mL/ vial	processed human plasma negative and positive for anti-HBc with preservatives	2–8°C	until the expiration date on the vial or onboard 8 hours
ADVIA Centaur WASH 1**	Wash 1	1500 mL/ pack	phosphate buffered saline with sodium azide (< 0.1%) and surfactant	2–25°C	until the expiration date on the vial or onboard 14 days

* The Chaotrope Reagent is colorless to light pink in color.

** See *Materials Required But Not Provided*

Precautions and Warnings

For *In Vitro* Diagnostic Use.

CAUTION: Sodium azide can react with copper and lead plumbing to form explosive metal azides. On disposal, flush reagents with a large volume of water to prevent the buildup of azides, if disposal into a drain is in compliance with federal, state, and local requirements.

R22
R32
S14
S28

Harmful! Harmful if swallowed. Contact with acids liberates very toxic gas. Keep away from acids. After contact with skin, wash immediately with plenty of soap and water. **Contains:** potassium thiocyanate; Chaotrope Reagent, Ancillary Reagent

R43
S24
S37

Irritant! May cause sensitization by skin contact. Avoid contact with skin. Wear suitable gloves. **Contains:** 5-chloro-2-methyl-2H-isothiazol-3-one and 2-methyl-2H-isothiazol-3-one; included in controls.

CAUTION! POTENTIAL BIOHAZARD: Some components of this product contain human source material. No known test method can offer complete assurance that products derived from human blood will not transmit infectious agents. All products manufactured using human source material should be handled as potentially infectious. Handle this product according to established good laboratory practices and universal precautions.[7-9]

The negative control has been assayed by FDA-approved methods and found nonreactive for hepatitis B surface antigen (HBsAg), antibody to hepatitis C (HCV), and antibody to HIV-1/2. The positive control and calibrators contain human plasma that may be reactive for HBsAg. The units were treated with a BPL-UV inactivation procedure, however, all products manufactured using human source material should be handled as potentially infectious.

Loading Reagents

Ensure that the system has sufficient primary and ancillary reagent packs. For detailed information about preparing the system, refer to the system operating instructions or to the online help system.

CAUTION: Mix all primary reagent packs by hand before loading them onto the system. Visually inspect the bottom of the reagent pack to ensure that all particles are dispersed and resuspended. For detailed information about preparing the reagents for use, refer to Appendix C, *Handling Reagents* in the ADVIA Centaur Assay Manual.

Load the ReadyPack primary reagent packs in the primary reagent compartment using the arrows on the packs as a placement guide. The system automatically mixes the primary reagent packs to maintain homogeneous suspension of the reagents. For detailed information about loading reagents, refer to the system operating instructions or to the online help system.

CAUTION: The Low and High Calibrators provided in this kit are matched to the ReadyPack primary reagent pack. Do not mix calibrator lots with different lots of reagent packs.

CAUTION: The Ancillary Reagent provided in this kit is matched to the Solid Phase and Lite Reagent. Do not mix Ancillary Reagent lots with different lots of Solid Phase and Lite Reagent.

Onboard Stability and Calibration Interval

Onboard Stability	Calibration Interval
28 days	14 days

Additionally, the ADVIA Centaur HBc Total assay requires a two-point calibration:

- when changing lot numbers of primary reagent packs
- when replacing system components
- when quality control results are repeatedly out of range

CAUTION:

- Discard reagent packs at the end of the onboard stability interval.
- Do not use reagents beyond the expiration date.

Master Curve Calibration

The ADVIA Centaur HBc Total assay requires a Master Curve calibration when using a new lot number of Lite Reagent, Solid Phase, and Chaotrope Reagent. For each new lot number of Lite Reagent, Solid Phase, and Chaotrope Reagent use the barcode reader or keyboard to enter the Master Curve values on the system. The Master Curve card contains the Master Curve values. For detailed information about entering calibration values, refer to the system operating instructions or to the online help system.

Calibration

For calibration of the ADVIA Centaur HBc Total assay, use ADVIA Centaur HBc Total Calibrators provided with each kit. The calibrators provided in this kit are matched to the ReadyPack primary reagent pack.

Using Barcode Labels

NOTE: Calibrator barcode labels are lot number specific. Do not use barcode labels from one lot of calibrators with any other lot of calibrators.

Use the ADVIA Centaur HBc Total Calibrator barcode labels to identify the Low and High Calibrator sample cups when performing the ADVIA Centaur HBc Total assay. Place the barcode label on the sample cup so that the readable characters on the side of the label are vertical on the sample cup.

Performing a Calibration

Each lot of calibrators contains a Calibrator Assigned Value card to facilitate entering the calibration values on the system. Enter the values using the barcode scanner or the keyboard. For detailed information about entering calibrator values, refer to the system operating instructions or to the online help system.

NOTE: This procedure uses calibrator volumes sufficient to measure each calibrator in duplicate.

1. Schedule the calibrators to the worklist.
2. Label two sample cups with calibrator barcode labels: one for the low and another for the high.

NOTE: Each drop from the calibrator vial is approximately 50 µL.

3. Gently mix the Low and High Calibrators and dispense at least 4 to 5 drops into the appropriate sample cups.
4. Load the sample cups in a rack.
5. Place the rack in the sample entry queue.
6. Ensure that the assay reagents are loaded.
7. Start the entry queue, if required.

NOTE: Dispose of any calibrator remaining in the sample cups after 8 hours. Do not refill sample cups when the contents are depleted; if required, dispense fresh calibrators.

Quality Control

For quality control of the ADVIA Centaur HBc Total assay, use ADVIA Centaur HBc Total quality control materials. Refer to the Expected Value card for the suggested expected values specific for the lot number of the positive and negative controls. Additional controls may be tested according to guidelines or requirements of local, state, and/or federal regulations or accrediting organizations.

NOTE: The quality control material furnished is intended to monitor substantial reagent failure. If additional controls are desired, it is recommended to run a negative control and a positive control close to the clinically relevant point. Further, the quality control material furnished is in a serum matrix. It may not adequately control the assay for plasma specimens. The user should provide alternate control material for plasma matrix.

Using Barcode Labels

NOTE: Control barcode labels are lot number specific. Do not use barcode labels from one lot of controls with any other lot of controls.

Use the ADVIA Centaur HBc Total quality control barcode labels to identify the positive and negative sample cups when performing the ADVIA Centaur HBc Total assay. Place the barcode label on the sample cup so that the readable characters on the side of the label are vertical on the sample cup.

Performing Quality Control

For detailed information about entering quality control values, refer to the system operating instructions or to the online help system.

To monitor system performance and chart trends, as a minimum requirement, quality control samples should be assayed on each workshift that samples are analyzed. Quality control samples should also be assayed when performing a two-point calibration. Treat all quality control samples the same as patient samples.

NOTE: This procedure uses control volumes sufficient to measure each control in duplicate.

1. Schedule the quality control samples to the worklist.
2. Label two sample cups with quality control barcode labels: one for the positive, and another for the negative.

NOTE: Each drop from the control vial is approximately 50 µL.

3. Gently mix the quality control materials and dispense at least 5 to 6 drops into the appropriate sample cups.
4. Load the sample cups in a rack.
5. Place the rack in the sample entry queue.
6. Ensure that the assay reagents are loaded.
7. Start the entry queue, if required.

NOTE: Dispose of any quality control materials remaining in the sample cups after 8 hours. Do not refill sample cups when the contents are depleted; if required, dispense fresh quality control materials.

Taking Corrective Action

If the quality control results do not fall within the suggested Expected Values or within the laboratory's established values, then do the following:

- consider the sample results invalid and repeat testing if controls are out of range
- Investigate and determine the cause for the unacceptable control results
- review these instructions to ensure that the assay was performed according to the procedures recommended by Bayer HealthCare

- verify that the materials are not expired
- verify that required maintenance was performed
- if necessary contact Bayer HealthCare for more assistance
- When the condition is corrected, retest the controls and confirm that results are within acceptable limits.
- It is advisable to repeat some or all patient specimens before reporting results for this run.

Sample Volume

This assay requires 50 µL of sample for a single determination. This volume does not include the unusable volume in the sample container or the additional volume required when performing duplicates or other tests on the same sample. For detailed information about determining the minimum required volume, refer to *Sample Volume Requirements* in the *ADVIA Centaur Reference Manual.*

Assay Procedure

For detailed procedural information, refer to the system operating instructions or to the online help system.

CAUTION: Do not load more than one size of sample container in each rack. The rack indicator must be positioned at the correct setting for the size of sample container.

1. Prepare the sample container for each sample, and place barcode labels on the sample containers, as required.
2. Load each sample container into a rack, ensuring that the barcode labels are clearly visible.
3. Place the racks in the entry queue.
4. Ensure that the assay reagents are loaded.
5. Start the entry queue, if required.

Procedural Notes

Disposal

Dispose of hazardous or biologically contaminated materials according to the practices of your institution. Discard all materials in a safe and acceptable manner, and in compliance with all federal, state, and local requirements.

Interpretation of Results

For detailed information about how the system calculates results, refer to the system operating instructions or to the online help system.

The system reports anti-HBc total results in Index Values and as reactive or nonreactive.

- Samples with a calculated value of less than 0.50 Index Value are considered non reactive for total antibodies to hepatitis B core antigen.

- Samples with initial results in the range from 0.50 to 0.99 Index Value require retest. Repeat the testing in duplicate. Samples which are repeatedly ≥ 0.50 Index Value (by at least two of the three results) will be considered reactive for HBc Total. Samples which are repeatedly < 0.50 Index Value (by at least two of the three results) will be considered nonreactive for HBc Total.
- Samples with a calculated value greater than or equal to 1.00 Index Value are considered reactive for total antibodies to hepatitis B core antigen.
- The cutoff for the ADVIA Centaur HBc Total assay was verified based on results of clinical agreement generated from clinical studies.
- Sample results are invalid and must be repeated if the controls are out of range.

CAUTION: Heparin has been shown to decrease the Index values in some HBc Total reactive samples relative to serum (See under Alternative Sample Types). Results obtained from heparin specimens near the cutoff should be repeated with a serum specimen or interpreted with caution.

Limitations

- The ADVIA Centaur HBc Total assay is limited to the detection of total antibodies to hepatitis B core antigen in human serum or plasma (potassium EDTA plasma, lithium or sodium heparinized plasma).
- The results from this or any other diagnostic kit should be used and interpreted only in the context of the overall clinical picture. A negative test result does not exclude the possibility of exposure to hepatitis B virus. Levels of anti-HBc may be undetectable both in early infection and late after infection.
- The calculated values for hepatitis B in a given specimen, as determined by assays from different manufacturers, can vary due to differences in assay methods and reagent specificity. The results reported by the laboratory to the physician must include the identity of the assay used. Values obtained with different assay methods cannot be used interchangeably. The reported antibody level cannot be correlated to an endpoint titer
- Assay performance characteristics have not been established for immunocompromised, immunosuppressed, infants, children, or adolescent patients.
- Assay performance characteristics have not been established when the ADVIA Centaur HBc Total assay is used in conjunction with other manufacturers' assay for specific HBV serological markers.
- The performance of the ADVIA Centaur HBc Total assay has not been established with cord blood, neonatal specimens, cadaver specimens, heat-inactivated specimens, or body fluids other than serum or plasma, such as saliva, urine, amniotic fluid, or pleural fluid.
- Do not use specimens with obvious microbial contamination.
- A reactive anti-HBc Total result does not exclude co-infection by another hepatitis virus.

Expected Results

The prospective study population for the ADVIA Centaur® HBc Total assay consisted of 2016 patients. Of these 2016 patients, 961 patients (47.67%) were from the high risk population, 844 patients (41.87%) were from the signs and symptoms population, and 211 patients (10.46%) were from the dialysis population. The prospective study population was 42.56% Caucasian, 26.54% Hispanic,. 23.31% Black, 3.26% Asian, and 4.30% from unknown or other ethnicity. The majority of patients were male (54.02% male and 45.98% female). The mean age was 45.9 years (range of 12 to 82 years). Patients in the prospective study population were from the following geographic regions: Florida (38.79%), Texas (33.19%), New York (19.69%), and California (8.33%).

The ADVIA Centaur® HBc Total initial test results for the prospective population for all sites combined by age group and gender are summarized in the following table.

Bayer ADVIA Centaur® HBc Total Assay
Distribution of High Risk, Signs and Symptoms, and Dialysis Population by Age Group and Gender
All Testing Sites

Age (years)	Gender	Reactive[a]		Nonreactive[b]		Total	
		N	%	N	%	N	%
0-9	Male	0	--	0	--	0	--
	Female	0	--	0	--	0	--
	Overall	0	--	0	--	0	--
10-19	Male	2	28.57	5	71.43	7	31.82
	Female	1	6.67	14	93.33	15	68.18
	Overall	3	13.64	19	86.36	22	100.00
20-29	Male	15	17.86	69	82.14	84	45.65
	Female	16	16.00	84	84.00	100	54.35
	Overall	31	16.85	153	83.15	184	100.00
30-39	Male	77	39.29	119	60.71	196	51.44
	Female	51	27.57	134	72.43	185	48.56
	Overall	128	33.60	253	66.40	381	100.00
40-49	Male	216	56.10	169	43.90	385	56.45
	Female	106	35.8	190	63.97	296	43.46
	Overall	322	47.28	359	52.64	681	100.00
50-59	Male	172	60.35	113	39.24	285	58.40
	Female	76	37.43	127	61.95	203	41.59
	Overall	248	50.82	240	48.68	488	100.00
60-69	Male	37	43.02	49	56.32	86	46.52
	Female	28	28.00	72	72.00	100	53.48
	Overall	65	34.95	121	64.71	186	100.00
>70	Male	21	46.67	24	53.33	45	61.64
	Female	5	17.86	23	82.14	28	38.36
	Overall	26	35.62	47	64.38	73	100.00
Unknown	Male	1	100.00	0	--	1	100.00
	Female	0	--	0	--	0	--
	Overall	1	100.00	0	--	1	100.00
Total	Male	541	49.67	548	50.14	1089	54.02
	Female	283	30.53	644	69.25	927	45.98
	Overall	824	40.87	1192	58.92	2016	100.00

a Samples with an Index Value ≥ 0.50
b Samples with an Index Value < 0.50

As with all in vitro diagnostic assays, each laboratory should determine its own reference range(s) for the diagnostic evaluation of patient results.[19]

Performance Characteristics

Results by Specimen Classification

The HBV disease classification for each patient in the high risk, signs and symptoms, and dialysis populations (2016 patients total) was determined by serological assessment using resultant hepatitis marker profiles obtained from results of commercially available, USFDA-approved reference assays. The serological assessment included the following 6 HBV markers: hepatitis B virus surface antigen (HBsAg), hepatitis B virus e antigen (HBeAg), total antibody to hepatitis B virus core antigen (Anti-HBc Total), IgM antibody to hepatitis B virus core antigen (Anti-HBc IgM), total antibody to HBeAg (Anti-HBe), and total antibody to hepatitis B virus surface antigen (Anti-HBs) (quantitative). Testing of these specimens occurred at each study site. The individual ADVIA Centaur® HBV assay result was compared to the reference HBV assay result and to the patient classification. No patients were excluded from the complete study set because of incomplete reference HBV serological results.

Each patient's HBV infection was classified based on the reactive (+)/nonreactive(-) patterns of the 6 HBV reference serological markers. Disease classification for each patient was based only on the HBV serological marker results, and was not affected by additional laboratory or clinical information. There were 31 unique reference marker patterns observed. These patterns are presented in the following table.

HBV Classification	HbsAg[a]	HBeAg	Anti-HBc IgM	Anti-HBc Total	Anti-HBe	Anti-HBs (>10mIU/mL)
Acute	+	+	+	+	+	-
Acute	+	+	+	+	-	-
Acute	+	-	+	+	+	-
Chronic	+	+	-	+	+	-
Chronic	+	+	-	+	-	+
Chronic	+	+	-	+	-	-
Chronic	+	-	-	+	+	+
Chronic	+	-	-	+	+	-
Chronic	+	-	-	+	-	+
Chronic	+	-	-	+	-	-
Chronic	+	+	+	+	-	+
Early Recovery	-	-	+	+	+	+
Early Recovery	-	-	+	+	+	-
Early Recovery	-	-	+	+	-	+
Early Recovery	-	-	+	+	-	-
Early Recovery	-	-	-	+	+	-
Recovery	-	-	-	+	+	+
Recovery	-	-	-	-	+	+
Recovered	-	-	-	+	-	+
Recovered	-	-	-	+	-	-
HBV Vaccine Response	-	-	-	-	-	+
Not previously infected	-	-	-	-	-	-
Uninterpretable	+	-	-	-	-	+
Uninterpretable	+	-	-	-	-	-
Uninterpretable	-	+	-	-	-	+
Uninterpretable	-	+	-	-	-	-
Uninterpretable	-	-	+	-	-	-
Uninterpretable	-	-	-	-	+	-
Uninterpretable	-	+	-	+	-	+
Uninterpretable	-	+	-	+	-	-
Uninterpretable	-	+	-	+	+	+

+ = reactive

- = nonreactive

(a) reactive (+) = reference HBsAg assay result was reactive and confirmed to be positive by neutralization nonreactive (-) = reference HBsAg assay results was nonreactive or reactive but not confirmed positive by neutralization.

Note: when the result was equivocal or indeterminate, it was assumed to be nonreactive (-) for classification purposes.

Comparison of Results

Following the assignment of specimen classification, the HBV results obtained using the ADVIA Centaur® method were compared with results obtained using the reference method for each result category (reactive and nonreactive). The method comparison for all testing sites combined is presented in the following table.

Method Comparison in High Risk, Signs and Symptoms, and Dialysis Population by HBV Classification
ADVIA Centaur® HBc Total Assay vs. anti-HBc Total Reference Assay
All Testing Sites

	Reference anti-HBc Total Negative		Reference anti-HBc Total Positive		Total[a]
	ADVIA Centaur® HBc Total Assay		ADVIA Centaur® HBc Total Assay		
HBV Classification	Reactive	Nonreactive	Reactive	Nonreactive	
	N	N	N	N	N
Acute	0	0	11	0	11
Chronic	0	0	111	1	112
Early Recovery	0	0	123	0	123
Recovery	1	1	208	1	211
Recovered	0	0	269	51	320
HBV Vaccine Response	15	368	0	0	383
Not Previously Infected	27	808	0	0	835
Uninterpretable	1	14	6	0	21
Total	44	1191	728	53	2016

a Total number of test results by HBV categories

Percent Agreement

The percent agreement between the ADVIA Centaur® HBc Total assay and the reference anti-HBc Total assay for the high risk, signs and symptoms, and dialysis populations across all testing sites is summarized in the following table.

Bayer ADVIA Centaur® HBc Total Assay
Percent Agreement and Confidence Intervals by HBV Classification in High Risk, Signs and Symptoms, and Dialysis Population
ADVIA Centaur® HBc Total Assay vs. anti-HBc Total Reference Assay
All Testing Sites

HBV Classification	Positive Percent Agreement % (x/n)[a]	95% Confidence Interval	Negative Percent Agreement % (x/n)[b]	95% Confidence Interval
Acute	100.00 (11/11)	71.51 to 100.00	--	--
Chronic	99.11 (111/112)	95.13 to 99.98	--	--
Early Recovery	100.00 (123/123)	97.05 to 100.00	--	--
Recovery	99.52 (208/209) *	97.36 to 99.99	50.00 (1/2)**	1.26 to 98.74
Recovered	84.06 (269/320)***	79.58-87.90	--	
HBV Vaccine Response	--		96.08 (368/383)	93.62-97.79
Not Previously Infected	--		96.77(808/835)	95.33-97.86
Uninterpretable	100.00 (6/6)	54.07 to 100.00	93.33 (14/15)	68.05 to 99.83
Overall	93.21 (728/781)	91.22 to 94.88	96.43 (1191/1235)	91.90 to 97.65

a *x = the number of ADVIA Centaur® HBc Total results that are reactive in agreement with the reference anti-HBc Total; n = the total number of reference anti-HBc Total results that are reactive*
b *x = the number of ADVIA Centaur® HBc Total results that are nonreactive in agreement with the reference anti-HBc Total; n = the total number of reference anti-HBc Total results that are nonreactive*

These samples were tested with a second FDA approved anti-HBc assay with the following results (see discussion below) :
**Recovery - 1 discrepant sample was reactive when tested with a 2nd FDA approved assay*
***Recovery - 1 discrepant sample was reactive when tested with a 2nd FDA approved assay*
****Recovered - 22 of 51 discrepant samples were non-reactive when tested with a 2nd FDA approved assay*

Comparison of Results, Retrospective Population

A retrospective study was conducted using 49 well characterized, commercially available samples from patients diagnosed with acute HBV and 104 well-characterized, commercially available samples from patient with chronic HBV (patients who had a positive HBsAg result at least 6 months prior to sample collection). Samples were evaluated using the ADVIA Centaur HBc Total assay and an anti-HBc Total reference assay. The method comparison for the acute and chronic retrospective population across all testing sites is presented in the following table. Positive percent agreement and negative percent agreement for this population were both 100.00%. The following results were obtained.

Bayer ADVIA Centaur® HBc Total Assay
Method Comparison in Retrospective[a] HBV Infected Population
ADVIA Centaur® HBc Total Assay vs. anti-HBc Total Reference Assay
All Testing Sites

	Reference anti-HBc Total Negative		Reference anti-HBc Total Positive		
	ADVIA Centaur® HBc Total Assay		ADVIA Centaur® HBc Total Assay		Total[b]
HBV classification	Reactive N	Nonreactive N	Reactive N	Nonreactive N	N
Acute	0	0	49	0	49
Chronic	0	0	104	0	104
Total	0	0	153	0	153

a Vendor assignment of acute specimens was based on positive HBsAg and Anti-HBcM test results. Assignment of chronic specimens was based on positive HBsAg test result 6 months after diagnosis of HBV. Vendor assignment was verified based on single replicate testing of HBsAg, Anti-HBc Total, and Anti-HBs assays at sites.

b Total number of test results by HBV categories

Seroconversion Panels

Commercially available HBV patient seroconversion panels were tested using the ADVIA Centaur HBc Total assay to determine the seroconversion sensitivity of the assay. The following results were obtained:

	Anti-HBc Total Positive Result From Initial Draw Date		Reference Assay vs ADVIA Centaur Assay
Panel ID	Reference Assay (Days)	ADVIA Centaur Assay (Days)	Difference in Bleed Numbers*
RP-009	29	29	0
RP-0016	60	57	+1
RP-0017	71	71	0
BCP-6281	41	41	0
Nabi-SB0413	62	62	0
Nabi-SB0411	35	35	0
Serologicals 22663D	63	63	0

* The difference in bleed numbers is relative to the reference assay. For example, a +1 means that the reference assay required 1 additional bleed before reactivity was determined as compared to the time-point when ADVIA Centaur assay confirmed positive.

Precision

Precision was evaluated according to the National Committee for Clinical Laboratory Standards protocol EP5-A.[11] A 6 member panel and controls were assayed in 2 replicates, 2 times a day, for 20 days. The following results were obtained using 1 reagent lot and a stored calibration curve.

	Mean Index Value	Within-run		Run-to-run		Total	
Sample		SD	CV (%)	SD	CV (%)	SD	CV (%)
Serum 1	0.06	0.02	NA*	0.01	NA	0.02	NA
Serum 2	1.13	0.06	5.1	0.05	4.5	0.09	8.4
Serum 3	1.22	0.04	3.6	0.05	4.2	0.08	6.8
Serum 4	1.35	0.08	5.8	0.05	3.8	0.10	7.1
Serum 5	2.40	0.12	4.9	0.08	3.4	0.19	7.7
Serum 6	4.54	0.20	4.4	0.21	4.7	0.29	6.5
Control Low	0.45	0.03	6.7	0.02	3.4	0.04	8.0
Control High	3.69	0.12	3.3	0.13	3.6	0.22	6.0

NA* = Not applicable

System Reproducibility

The ADVIA Centaur HBc Total reproducibility study was performed at 3 external sites using 2 reagent lots per site. A twenty member panel and controls were assayed in replicates of 5 on a single run per day over 6 days for each lot. The study was completed within a single calibration of the assay (one calibration interval). The maximum number of replicates used was 180. Control Lot 782174 was used at two sites and the reproducibility analyses for this lot included 120 replicates. A second Control lot, 783154, was used at only one site and the reproducibility analyses for this lot included 60 replicates. Replicates of negative samples (Panel member 1) reported as below the reportable range were non-numerical results and were excluded from the analyses. Eight positive panel member results were determined to be outliers and were excluded from the analyses.

The data from all 3 sites and from all 3 reagent lots were combined to obtain SD and percent CV for within run, between run, between testing site, between reagent lot, and total. The precision estimates were derived from variance component analysis. A NESTED SAS model was used for analysis. The reproducibility results are presented in the following table:

Bayer ADVIA Centaur HBcT Index Reproducibility between Reagent Lots and Testing Sites

			Within Run [a]		Between Run [b]		Between Site [c]		Between Reagent Lots [d]		Total [e]		
Panel member or Control Level	Matrix or Control Lot	Mean Index Value	SD	CV %	SD	CV %	SD	CV %	SD	CV %	SD	CV %	Number of Observations
1	EDTA	0.10	0.01	NA	0.01	NA	0.04	NA	0.03	NA	0.05	NA	177
1	Li Heparin	0.09	0.02	NA	0.01	NA	0.03	NA	0.02	NA	0.04	NA	156
1	Na Heparin	0.08	0.01	NA	0.00	NA	0.02	NA	0.03	NA	0.04	NA	150
1	Serum	0.08	0.02	NA	0.01	NA	0.02	NA	0.03	NA	0.04	NA	158
2	EDTA	0.71	0.04	6.2	0.04	5.8	0.05	6.7	0.02	2.8	0.08	11.2	180
2	Li Heparin	0.69	0.04	6.5	0.05	6.6	0.03	5.0	0.04	5.3	0.08	11.8	180
2	Na Heparin	0.64	0.05	7.9	0.05	7.5	0.06	9.9	0.00	0.0	0.09	14.8	180
2	Serum	0.68	0.04	6.4	0.06	8.3	0.02	3.0	0.05	7.2	0.09	13.1	180
3	EDTA	0.77	0.05	6.5	0.06	7.3	0.03	4.2	0.03	4.4	0.09	11.5	180
3	Li Heparin	0.75	0.05	7.3	0.06	7.4	0.03	4.2	0.03	3.4	0.09	11.7	175
3	Na Heparin	0.70	0.05	7.1	0.03	4.9	0.06	9.1	0.00	0.0	0.09	12.5	180
3	Serum	0.75	0.05	6.1	0.06	8.7	0.02	3.1	0.04	5.0	0.09	12.1	180
4	EDTA	1.09	0.08	7.7	0.07	6.7	0.07	6.8	0.06	5.3	0.15	13.4	180
4	Li Heparin	1.03	0.07	6.8	0.05	5.0	0.08	7.6	0.07	6.8	0.14	13.2	180
4	Na Heparin	0.98	0.07	6.8	0.07	6.9	0.08	7.8	0.00	0.0	0.12	12.5	180
4	Serum	1.07	0.07	6.6	0.11	10.3	0.00	0.0	0.05	4.7	0.14	13.1	180
5	EDTA	2.75	0.16	6.0	0.20	7.4	0.14	5.0	0.17	6.0	0.34	12.3	179
5	Li Heparin	2.73	0.15	5.7	0.15	5.6	0.17	6.3	0.15	5.6	0.32	11.6	180
5	Na Heparin	2.57	0.12	4.8	0.15	5.8	0.15	6.0	0.00	0.0	0.25	9.6	178
5	Serum	2.78	0.19	6.9	0.22	8.0	0.10	3.4	0.14	5.1	0.34	12.2	180
Low Control	782174	0.39	0.03	8.3	0.02	5.6	0.06	15.7	0.00	0.0	0.07	18.7	120
Low Control	783154	0.40	0.03	6.5	0.01	2.9	NA	NA	0.03	7.0	0.04	10.0	60
High Control	782174	2.86	0.19	6.5	0.11	4.0	0.15	5.3	0.00	0.0	0.27	9.3	120
High Control	783154	3.06	0.12	4.0	0.13	4.3	NA	NA	0.00	0.0	0.18	5.8	60

SD = standard deviation

CV = coefficient of variation

NA = not applicable

[a] Variability of the assay performance from replicate to replicate

[b] Variability of the assay performance from day to day

[c] Variability of the assay performance from site to site

[d] Variability of the assay performance from reagent lot to reagent lot

[e] Variability of the assay performance combining the effect of all four components

Analytical Sensitivity

To examine the analytical sensitivity of the ADVIA Centaur HBc Total assay the Paul Ehrlich Institute (PEI) anti-HBc Total reference sample was used to prepare a dilution series which was assayed using three ADVIA Centaur HBc Total reagent lots. Linear regression was used to determine the concentration of PEI reference sample which corresponds to the ADVIA Centaur HBc Total cutoff (Index Value = 0.5). The PEI International Unit (IU) concentration at the assay cutoff was determined to be 0.2 PEI IU/mL.

Cross-Reactivity

The ADVIA Centaur HBc Total assay was evaluated for potential cross-reactivity with other viral antibodies and disease state specimens. The anti-HBc Total status of each specimen was verified using an anti-HBc Total reference assay. The following results were obtained using the ADVIA Centaur HBc Total assay.

		Number of Positive Anti-HBc Total Results	
Clinical Category	Number Tested	ADVIA Centaur Assay	Reference Assay
Hepatitis A Infection (HAV)	5	1	1
Hepatitis C Infection (HCV)	10	4	4
Epstein-Barr Virus (EBV) IgG	10	2	2
Epstein-Barr Virus (EBV) IgM	10	3	3
Herpes Simplex Virus (HSV) IgG	10	3	3
Herpes Simplex Virus (HSV) IgM	10	4	4
Cytomegalovirus IgG	10	7	7
Cytomegalovirus IgM	3	1	1
Toxoplasma IgG	10	2	2
Toxoplasma IgM	7	0	0
Syphilis IgG	10	1	1
Human Immunodeficiency Virus (HIV1/2)	10	2	2
Varicella Zoster IgG	10	4	4
Rubeola IgG	10	5	4[1]
Non viral Liver Disease	9	0	0
Autoimmune Disease (Rheumatoid Arthritis)	9	0	0
Anti-Nuclear Antibody (ANA)	5	1	1
Systemic Lupus Erythematosus (SLE)	2	0	0
HAMA	10	0	0
Flu vaccine Recipient	10	3	3
Total Samples Tested	170	43	42

1. The non-confirmed ADVIA Centaur HBc Total reactive result was ADVIA Centaur anti-HBs positive.

Endogenous Interferents

The potentially interfering effects of conjugated bilirubin, unconjugated bilirubin, hemoglobin, triglycerides, hyper IgG and low protein were evaluated following the guidelines described by NCCLS EP7-P[12] for interference due to endogenous substances.

Serum specimens that are . . .	*Demonstrate ≤10% change in results up to . . .*
hemolyzed	500 mg/dL of hemoglobin
lipemic	1000 mg/dL of intralipids
icteric	60 mg/dL of conjugated bilirubin
icteric	40 mg/dL of unconjugated bilirubin
proteinemic	12.0 g/dL of protein
proteinemic	3.5 g/dL of protein
Hyper IgG	60 mg/mL of immunoglobulin G

Alternative Sample Types

The ADVIA Centaur HBc Total assay can use plasma specimens collected using either potassium EDTA, sodium heparin, or lithium heparin anticoagulants. In a matched study of 217 serum, potassium EDTA plasma and lithium heparin plasma samples the plasma samples had Index Values equivalent to those of the serum samples collected from the same patients. Results of the paired matrix study are summarized in the table below.

Bayer ADVIA Centaur® HBc Total Assay Matrix Study Summary (All Testing Sites)	
Specimen Type	***Mean ADVIA Centaur® HBc Total Index Value***
Serum (control)	*2.65*
EDTA plasma	*2.63*
Lithium heparin plasma	*2.66*
Difference in Mean ADVIA Centaur® HBc Total Index Value	
Control vs. EDTA	*Control vs. Heparin*
0.03 (*P* = 0.0262)[a]	*-0.01 (P = 0.1297)*[a]

a P value for the comparison of difference of Mean Serum Control Index versus Mean Heparin/Mean EDTA Index (2-sample comparison: t-Test or Wilcoxon Rand Test as appropriate after testing for normality).

Technical Assistance

For customer support, please contact your local technical support provider or distributor.

References

1. Gitlin N. Hepatitis B: Diagnosis, Prevention, and Treatment. Clinical Chemistry 1997, 43:8(B):1500-1506.
2. Mahoney, FJ. Update on Diagnosis, Management, and Prevention of Hepatitis B Virus Infection. Clinical Microbiology Reviews 1999,12(2):351-366.
3. Juszczyk, J. Clinical Course and Consequences of Hepatitis B infection. Vaccine 2000, 18:S23-S25.
4. Vivek R. Treatment of hepatitis B. Clin Cornerstone 2001, 3(6):24-36.
5. Koff RS. Hepatitis B today: Clinical Diagnostic Overview. The Pediatric Infectious Disease Journal 1993 May: 12(5): 428-32.
6. National Committee for Clinical Laboratory Standards. Procedures for handling and processing of blood specimens; approved guideline. NCCLS Document H18-A2. Wayne (PA): NCCLS; 1999 Oct. 40p.
7. Centers for Disease Control. Update: Universal precautions for prevention of transmission of human immunodeficiency virus, hepatitis B virus and other bloodborne pathogens in healthcare settings. MMWR 1988;37:377-82, 387-8.
8. National Committee for Clinical Laboratory Standards. Protection of laboratory workers from instrument biohazards and infectious disease transmitted by blood, body fluids, and tissue; approved guideline. NCCLS Document M29-A2. Wayne (PA):NCCLS; 2001 Dec. 105p.
9. Federal Occupational Safety and Health Administration, Bloodborne Pathogens Standard, 29 CFR 1910.1030.
10. National Committee for Clinical Laboratory Standards. How to define, determine, and utilize reference intervals in the clinical laboratory; approved guideline. NCCLS Document C28-A. Wayne (PA):NCCLS;1995.
11. National Committee for Clinical Laboratory Standards. Evaluation of precision performance of clinical chemistry devices–second edition; approved guideline. NCCLS Document EP5-A. Wayne (PA):NCCLS;1999.
12. National Committee for Clinical Laboratory Standards. Interference Testing in Clinical Chemistry; Proposed Guidelines. NCCLS document EP7-P. Wayne (PA): NCCLS: 1986